



New culturing tool reveals a full genome from single cells

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Gel microdroplet culturing reveals intraspecies genomic diversity within the human microbiome

LOS ALAMOS, N. M., March 15, 2013—A new technique for genetic analysis, “gel microdroplets,” helps scientists generate complete genomes from a single cell, thus opening the door to understanding the complex interrelationships of bacteria, viruses and eukaryotes that form “microbiome” communities in soil, in humans, and elsewhere in the natural world.

Microbes live in complex communities that function together as a whole in order to survive and thrive in their natural environments. Microbes survive almost everywhere, and they make up the majority of the living organisms on Earth and contribute to all aspects of human life, such as health, energy and even climate change.

Most types of bacteria cannot grow in the laboratory as a pure, isolated culture, however, due to complicated interactions that support their growth. This makes research challenging, as identifying a single organism’s genetic profile fails to take into account the interrelationships that are extremely important to understanding the microbe’s roles and capabilities in its specific location.

Scientists from Los Alamos National Laboratory and the J. Craig Venter Institute in San Diego have made a breakthrough that gives researchers the bigger picture of the multi-organism genome, using the complete genome from a single cell.

The technique used over the past few years, metagenomics, avoids the need for culturing to produce mixed genetic info for the whole community. However, many of the biological questions, such as how the mixed bacterial or viral community members interact with each other, cannot be answered without genomic information about the various individual species in the community. The Los Alamos group, led by Cliff Han, Michael Fitzsimons (formerly of LANL), and Armand Dichosa, has been developing technologies to fulfill the need.

The technology has the potential to generate complete genomes from single cells of traditionally uncultured species. Using gel microdroplets (GMD), the science team created dozens to hundreds of identical cells from single cells, while keeping such cells separated from the rest of community and maintaining the cells’ ability to communicate with other community members.

From mixed bacterial communities inhabiting the human mouth and digestive gut, researchers captured single cells within microscopic GMD and incubated them in a defined growth medium.

The characteristic pores and channels of the agarose-based GMD allow for the movement of nutrients, chemical signals and metabolic wastes to and from the living cell as if it were in its natural environment. The captured single cells multiply to microcolonies of hundreds, thereby producing sufficient quantities of identical genomic templates. Ultimately, this allows for the completion of several genomes from the same bacterial species.

By completing and comparing the genomic profiles of these species, researchers found significant variations within the genomes of the same orally-located species, with few differences found from within gut-resident species. Such findings show how significantly active (or inactive) bacteria are in recombining specific segments of DNA with each other and raise questions as to how we identify a “species” if something as important as its neighborhood interactions can change its genetic profile.

With promising results of this human microbiome study, the team has begun to use GMD to culture bacteria and archaea in their native environments, such as wetland and water environments. Researchers want to capture known, rare and elusive species that cannot grow in laboratory settings, and also to provide completed genomes of these novel species that may, again, offer insight into the vital contributions of bacteria and archaea in local ecology and global climate change.

“We have demonstrated a novel approach for fully sequencing genomes of microorganisms found in complex communities,” said Dichosa.

Previously, complete community genomes had been an unattainable goal because neither of the two competing technologies, shotgun metagenomics or single-cell sequencing, can recover a nearly complete genome from a single organism in a diverse sample. “We believe using GMDs to sequence complete genomes from environmental samples shows great promise and will allow for the first time a high throughput technology for exploring community pan-genomics,” said Han.

The research team’s paper, “Nearly finished genomes produced using gel microdroplet culturing reveal substantial intraspecies genomic diversity within the human microbiome.” is published in the online advance March 14 edition of [Genome Research](#).

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